

Claims

- 1 1. A polynucleotide comprising:
2 a nucleotide sequence encoding an RNAi operatively linked to a tissue specific
3 promoter, a cell specific promoter, and/or an inducible promoter.
- 1 2. The polynucleotide according to claim 1, wherein the promoter is a tissue specific
2 promoter or a cell specific promoter.
- 1 3. The polynucleotide according to claim 1 or claim 2, wherein the promoter is a
2 tissue specific and/or cell specific and/or inducible RNA polymerase II promoter.
- 1 4. The polynucleotide according to any one of claims 1 to 3, wherein the promoter is
2 selected from the group consisting of a collagen 1A1 promoter, a collagen 1A2
3 promoter, a collagen 3A1 promoter, a GNAT-2 promoter, a peripherin-rds
4 promoter, a rhodopsin promoter, a cone arrestin promoter, a RPE65 promoter, an
5 albumin promoter, an insulin promoter, a presenillin 1 promoter, a presenillin 2
6 promoter, a Huntington's promoter, a super oxide dismutase promoter and an
7 enolase promoter.
- 1 5. The polynucleotide according to any one of claims 1 to 4, wherein the
2 polynucleotide further comprises at least one cleaving element that is capable of
3 cleaving a nucleotide sequence 5' and/or 3' to the RNAi sequence.
- 1 6. The polynucleotide according to claim 5, wherein the cleaving element is a
2 ribozyme, a maxizyme, or a minizyme.
- 1 7. The polynucleotide according to claim 7 wherein the cleaving element is a
2 ribozyme.
- 1 8. The polynucleotide according to any one of the preceding claims wherein the
2 cleaving element is operatively linked to a tissue specific promoter and/or a cell
3 specific promoter and/or an inducible promoter.

- 1 9. The polynucleotide according to claim 8 wherein the promoter to which the
2 cleaving element is operatively linked is selected from the group consisting of a
3 collagen 1A1 promoter, a collagen 1A2 promoter, a GNAT-2 promoter, a
4 peripherin-rds promoter, a rhodopsin promoter, an albumin promoter, an insulin
5 promoter, a presenillin 1 promoter, a presenillin 2 promoter, a super oxide
6 dismutase promoter, a Huntington's promoter and an enolase promoter.
- 1 10. The polynucleotide according to claim 8 or claim 9, wherein the promoter to
2 which the cleaving element is operatively linked is the same promoter to which
3 the nucleotide sequence encoding the RNAi is operatively linked.
- 1 11. The polynucleotide according to claim 8 or claim 9 wherein the promoter to
2 which the cleaving element is operatively linked is a different promoter to which
3 the nucleotide sequence encoding the RNAi is operatively linked.
- 1 12. The polynucleotide according to any one of claims 8 to 11, wherein the cleaving
2 element is cis-acting.
- 1 13. The polynucleotide according to any one of claims 8 to 12, wherein the
2 polynucleotide further comprises at least one suppression agent (e.g., a trans-
3 acting ribozyme) capable of suppression of one or more target gene(s) or
4 nucleotide sequences.
- 1 14. The polynucleotide according to any one of the preceding claims wherein the
2 polynucleotide further comprises a transcription termination sequence.
- 1 15. The polynucleotide according to any one of the preceding claims , wherein the
2 RNAi sequence is placed at or close to at least one TSS of the promoter.
- 1 16. The polynucleotide according to any one of the preceding claims wherein the
2 RNAi sequence comprises a region complementary or essentially complementary
3 to a target gene.

- 1 17. The polynucleotide according to any one of the preceding claims wherein the
2 RNAi sequence is capable of discriminating between different alleles of the same
3 gene.
- 1 18. A vector comprising a polynucleotide according to any one of the preceding
2 claims.
- 1 19. The vector according to claim 16, wherein the vector is selected from the group
2 consisting of an adenoviral vector, an adenoassociated viral vector, a retroviral-C
3 type vector such as MLV; a lentiviral vector such as HIV or SIV, herpes simplex
4 (HSV), and SV40.
- 1 20. A host cell comprising the polynucleotide according to any one of claims 1 to 17
2 or the vector according to any one of claims 18 and 19.
- 1 21. The host cell according to claim 20, wherein the polynucleotide is integrated into
2 the host cell genome.
- 1 22. The host cell according to claim 21, wherein the cell is selected from the group
2 consisting of a 661W cell line cell, a Y79 cell line cell, an MPCs cell, a
3 hepatocyte, an osteoblast, and a neuronal cell line cell.
- 1 23. A transgenic animal comprising the polynucleotide according to any one of
2 claims 1 to 17, the vector according to any one of claims 18 and 19 or the host
3 cell according to any one of claims 20 to 22.
- 1 24. A method of inhibiting or reducing expression of a target gene in a cell of an
2 organism, said method comprising the steps:
3 (i) administering to said cell a polynucleotide according to any one of claims 1 to
4 17 or the vector according to any one of claims 18 and 19, wherein the RNAi has
5 specificity or partial specificity for the target gene; and
6 (ii) allowing expression of the RNAi such that the RNAi inhibits or reduces
7 expression of the target gene.

- 1 25. The method according to claim 24, wherein said polynucleotide is integrated into
2 the genome of the cell.
- 1 26. A method of identifying a modulator of a target gene, said method comprising the
2 steps:
3 providing a host cell according to any one of claims 20 to 22 or a transgenic
4 organism according to claim 23,
5 (ii) administering a candidate modulator to said host cell or said transgenic
6 organism; and
7 (iii) determining expression of said target gene in the presence of the candidate
8 modulator.
- 1 27. The method according to claim 26, wherein the target gene is a disease gene
2 and/or predisposes an individual to a disease pathology and/or makes an
3 individual more susceptible to an infectious disease.
- 1 28. A pharmaceutical composition comprising a polynucleotide according to any one
2 of claims 1 to 17, the vector according to any one of claims 18 and 19 or the host
3 cell according to any one of claims 20 to 22 and a pharmaceutical excipient.
- 1 29. A polynucleotide according to any one of claims 1 to 17, the vector according to
2 any one of claims 18 and 19 or the host cell according to any one of claims 20 to
3 22 for use in a method of medical treatment or diagnosis.
- 1 30. Use of a polynucleotide according to any one of claims 1 to 17, the vector
2 according to any one of claims 18 and 19 or the host cell according to any one of
3 claims 20 to 22 in the preparation of a medicament for the treatment of retinitis
4 pigmentosa, epidermolysis bullosa, osteogenesis imperfecta, Ehlers-Danlos
5 syndrome, Marfan's disease, dominant negative cancers, Alzheimer's disease,
6 motor neuron disease, poly cystic kidney disease, or a disorder due to poly
7 glutamine expansions such as Huntington's chorea.

8 31. A method for establishing the biological function(s) of a target gene(s) in one or
9 more specific tissues and/or cell types according to any one of claims 20 to 22 or
10 a transgenic animal according to claim 23.